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Kerr, Janet
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Thanks,
Janet M. Kerr
A.U. 1633
305-4055
CM1-12A03

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L22 ANSWER 9 OF 11 CAPLUS COPYRIGHT 2001 ACS
AN 1996:381247 CAPLUS
DN 125:51280
TI **Screening** for reproductive **toxicity** in Fundulus heteroclitus by genetic expression **profiling**
AU Craig, J. C.; Westerman, M. E.; Bennett, G. D.; DiMichele, L.; Finnell, R. H.
CS Dep. Veterinary Anatomy Public Health, Texas A&M Univ., College Station, TX, 77843, USA
SO Biomarkers (1996), 1(2), 123-135
CODEN: BIOMFA; ISSN: 1354-750X
DT Journal
LA English
AB Potentially **teratogenic** agents enter the environment at a rate that greatly exceeds current capabilities to effectively evaluate their reproductive **toxicities**. This is due, in part, to costly, labor-intensive methodologies involving mammalian **embryonic screening assays** that are currently in use worldwide. Therefore, we sought to develop a rapid, less expensive **screening** system with which to identify mol. biomarkers of **teratogenicity** using a non-mammalian system. Embryos of the topminnow, Fundulus heteroclitus, offer several advantages in terms of reproductive **toxicity screening** efficiency as compared with mammalian **embryonic** systems. These embryos are easily manipulated and develop normally at ambient temp. in air, water, or air-satd. mineral oils, making them readily adapted for field studies. In the present study, developing F. heteroclitus embryos were exposed to **teratogenic** concns. of sodium valproate (VPA) or arsenic acid (arsenate), and the frequency and types of induced malformations were evaluated. Using in situ transcription and antisense RNA (aRNA) amplification procedures (IST/aRNA), we attempted to correlate the **teratogenic** outcomes to specific alterations in the expression of a panel of developmentally regulated genes. Preliminary studies identified treatment concns. of arsenate and VPA that induced abnormal development in 95% of the surviving embryos. Among the F. heteroclitus embryos, the structural defects most commonly induced by these compds. were cardiac and neural tube malformations. The genetic expression **profiles** revealed a no. of genes whose expression levels were significantly altered by exposure to the **test** compds. Mol. anal. of F. heteroclitus **embryonic** development represents a novel, inexpensive approach to **screen** for potential **teratogens**, and identify genes whose expression patterns may be used as biomarkers, or indicators, of **teratogenicity**.

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L22 ANSWER 11 OF 11 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1989:332748 BIOSIS
DN BA88:35748
TI THE USE OF ANIMAL MODELS IN UNDERSTANDING HUMAN **TERATOGENS**.
AU WEBSTER W S
CS DEP. ANAT., UNIV. SYDNEY, SYDNEY, N.S.W. 2006, AUST.
SO CONGENITAL ANOM, (1989) 28 (4), 295-302.
CODEN: CGANE7. ISSN: 0914-3505.
FS BA; OLD
LA English
AB The **testing** of **drugs** and other chemicals in pregnant animals is required by legislation in a number of countries as a **screening** procedure for **teratogenic** potenital in the human. The **testing** procedure involves methodology designed in the 1960s which was based on regimens established in the 1940s for **toxicity testing**. The requirement that animals are dosed to maternally **toxic** levels, frequently mean that the embryos are exposed to inappropriately high concentrations of the **test** substance. Positive results in this type of experiment may have no relevance to the human situation where the exposure **profile** is often quite different, with the human embryo being exposed for prolonged periods to much lower **drug** concentrations. One way of duplicating the anticipated human exposure is to grow rat embryos in serum containing the **drug** and/or its metabolites at concentrations determined in the human during early clinical **testing**. It is proposed that mammalian embryos will respond in a similar manner to a particular concentration of a **test** substance. In vitro experiments using isotretinoin and its main metabolite 4-oxo-isotretinoin showed that the metabolite was **teratogenic** at concentrations which occurred in the human during normal repetitive dosing and hence the metabolite was the likely human **teratogen**. Similarly, rat embryo culture studies showed that the anticonvulsant **drug**, valproic acid, was **teratogenic** at blood concentrations which occurred during normal dosing in the human. Other in vitro studies showed that cadmium is unlikely to be a human **teratogen**, despite the fact that is is well established as a **teratogen** in experimental animals in vivo. It is proposed that embryo culture should be used as an adjunct procedure during tertology **testing** making use of metabolic and pharmacokinetic data obtained from the human during clinical **testing**.

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L4 ANSWER 23 OF 25 MEDLINE
AN 94003746 MEDLINE
DN 94003746 PubMed ID: 8400633
TI Regulation of growth and differentiation in early development: of mice and models.
AU Mummery C L; Slager H G; van Inzen W; Freund E; van den Eijnden-Van Raaij A J
CS Hubrecht Laboratory, Netherlands Institute for Developmental Biology, Utrecht.
SO REPRODUCTIVE TOXICOLOGY, (1993) 7 Suppl 1 145-54. Ref: 66
Journal code: BE4; 8803591. ISSN: 0890-6238.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LA English
FS Priority Journals
EM 199311
ED Entered STN: 19940117
Last Updated on STN: 19940117
Entered Medline: 19931123
AB In this article we describe some of the fundamental processes occurring during early murine development, introduce cellular models used to investigate these processes and review some well-known factors that may be involved in their control. These include transforming growth factor beta, retinoic acid and leukaemia inhibitory factor. Refinements to the culture conditions of **embryonic stem** and embryonal carcinoma cells have enabled us to test the effects of these factors on growth and differentiation and in particular to establish that their interaction may determine the ultimate developmental state of the cell population. Preliminary studies using neutralizing antibodies in embryos are described that suggest that deregulation of normal **expression** can lead to a failure to implant. Insights into the events underlying normal embryonic development and implantation, yielded by the type of study described here, may contribute to an understanding of the mechanisms causing early embryonic loss and the role of **toxicants** in this process.

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L2 ANSWER 11 OF 11 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 6
AN 97166830 EMBASE
DN 1997166830
TI The **embryonic stem cell test**, an in vitro embryotoxicity test using two permanent mouse cell lines: 3T3 fibroblasts and embryonic stem cells.
AU Spielmann H.; Pohl I.; Doring B.; Liebsch M.; Moldenhauer F.
CS Dr. H. Spielmann, ZEBET, BgVV, Diedersdorfer Weg 1, D-12277 Berlin, Germany
SO In Vitro Toxicology: Journal of Molecular and Cellular Toxicology, (1997) 10/1 (119-127).
Refs: 17
ISSN: 0888-319X CODEN: IVTOE4
CY United States
DT Journal; Conference Article
FS 001 Anatomy, Anthropology, Embryology and Histology
021 Developmental Biology and Teratology
052 Toxicology
LA English
SL English
AB The **embryonic stem cell test** (EST) was developed as a new in vitro embryotoxicity test that does not use embryonic tissues from pregnant animals but only two permanent mouse cell lines: 3T3 fibroblasts and embryonic stem (ES) cells of the D3 line. In the EST, cytotoxicity was determined in the two cell lines for different time periods up to 10 days and, in addition, the differentiation of ES cells into contracting myocardial cells. Sixteen carefully selected test chemicals with different embryotoxic properties were tested in the EST. Of 12 endpoints and ratios of endpoints determined in the EST with the two cell lines, three endpoints were selected by stepwise discriminant analysis that showed a better correlation to the embryotoxic properties of the test chemicals than the other endpoints. Using the three endpoints and linear discriminant functions, a classification scheme was developed for the EST in which test chemicals are assigned to three classes of in vivo embryotoxicity: not embryotoxic, moderate and strong embryotoxic. Using this classification model all 16 test chemicals were correctly assigned in the EST to their in vivo classes of embryotoxicity. Such a promising result is usually not obtained in in vitro embryotoxicity tests, most of which are still using embryonic tissues taken from pregnant animals rather than permanent cell lines in the EST. The EST is, therefore, ready to undergo validation in other laboratories.

L2 ANSWER 8 OF 11 MEDLINE DUPLICATE 5
AN 2000075317 MEDLINE
DN 20075317 PubMed ID: 10592392
TI Embryotoxicity screening using embryonic stem cells in vitro: correlation to in vivo teratogenicity.
AU Scholz G; Pohl I; Genschow E; Klemm M; Spielmann H
CS Center for Documentation and Evaluation of Alternative Methods to Animal Experiments (ZEBET), Berlin, Germany.. zebet@bgvv.de
SO CELLS TISSUES ORGANS, (1999) 165 (3-4) 203-11. Ref: 38
Journal code: DCO; 100883360. ISSN: 1422-6405.
CY Switzerland
DT Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LA English
FS Priority Journals
EM 200002
ED Entered STN: 20000218
Last Updated on STN: 20000218
Entered Medline: 20000204
AB Blastocyst-derived pluripotent embryonic stem (ES) cells of the mouse can be induced to differentiate in culture into a variety of cell types, including cardiac muscle cells. The **embryonic stem cell test** that makes use of the differentiation of ES cells into cardiomyocytes in a standardized in vitro model was developed to offer an alternative method to comprehensive in vivo studies in reproductive toxicology about toxic effects of chemicals. ES cells of the mouse cell line D3 are investigated for their preserved capability to differentiate following drug exposure, and both ES cells and differentiated fibroblast cells of the mouse cell line 3T3 are comparatively analyzed for effects on viability. The following endpoints are used to classify the embryotoxic potential of chemicals into three classes of in vitro embryotoxicity (non-, weakly or strongly embryotoxic).
These endpoints are: (1) the inhibition of differentiation of ES cells into cardiomyocytes after 10 days of treatment, and the decrease of viability (cytotoxicity) of (2) 3T3 cells and (3) ES cells after 10 days of treatment, determined by a 3-(4,5-dimethylthiazol-2yl)-2,5-diphenyl tetrazolium bromide (MTT) test. 50% inhibition concentrations for differentiation (ID(50)) and cytotoxicity (IC(50)D3 and IC(50)3T3) are calculated from concentration-response curves. Applying linear analysis of discriminance, a biostatistical prediction model (PM) was developed. This procedure identified three variables, the lg(IC(50)D3), the lg(IC(50)3T3) and the relative distance between IC(50)3T3 and ID(50), that improved the separation of the three classes of embryotoxicity compared to the prediction model that was originally proposed after test development. Unlike the original PM, the improved PM incorporates as one variable the relative distance between IC(50)3T3 and ID(50), instead of the ratio ID(50)/IC(50)D3 that was used previously. Copyright Copyright 1999 S. Karger AG, Basel

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L4 ANSWER 2 OF 3 MEDLINE DUPLICATE 1
AN 2000394410 MEDLINE
DN 20362108 PubMed ID: 10900407
TI Development of prediction models for three in vitro embryotoxicity tests in an ECVAM validation study.
AU Genschow E; Scholz G; Brown N; Piersma A; Brady M; Clemann N; Huuskonen H;
Paillard F; Bremer S; Becker K; Spielmann H
CS Federal Institute for Health Protection of Consumers and Veterinary Medicine (BgVV), Berlin, Germany.
SO IN VITRO & MOLECULAR TOXICOLOGY, (2000 Spring) 13 (1) 51-66.
Journal code: DP4; 9808800. ISSN: 1097-9336.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200008
ED Entered STN: 20000824
Last Updated on STN: 20000824
Entered Medline: 20000815
AB Since 1997 the National Center for Documentation and Evaluation of Alternative Methods to Animal Experiments, **ZEBET**, in Berlin, has been coordinating a validation study aimed at prevalidation and validation of three in vitro embryotoxicity tests, funded by the European Center for the Validation of Alternative Methods (ECVAM) at the Joint Research Center (JRC, Ispra, Italy). The tests use the cultivation of postimplantation rat whole embryos (WEC test), cultures of primary limb bud cells of rat embryos (micromass or, MM, test), and cultures of a pluripotent mouse embryonic **stem** cell line (embryonic **stem** cell test or EST). Each of the tests was performed in four laboratories under blind conditions. In the preliminary phase of the validation study 6 out of 20 test chemicals comprising different embryotoxic potential (non, weakly, and strongly embryotoxic) were tested. The results were used to define biostatistically based prediction models (PMs) to identify the embryotoxic potential of test chemicals for the WEC test and the MM test. The PMs developed with the results of the preliminary phase of the validation study (training set) will be evaluated with the results of the remaining 14 test chemicals (definitive phase) by the end of the study. In addition, the existing, improved PM (iPM) for the EST, which had been defined previously, was evaluated using the results of the preliminary phase of this study. Applying the iPM of the EST to the results of this study, in 79% of the experiments, chemicals were classified correctly according to the embryotoxic potential defined by in vivo testing. For the MM and the WEC test, the PMs developed during the preliminary phase of this validation study provided 81% (MM test) and 72% (WEC test) correct classifications. Because the PM of the WEC test took into account only parameters of growth and development, but not cytotoxicity data, a second PM (PM2) was developed for the WEC test by incorporating cytotoxicity data of the differentiated mouse fibroblast cell line 3T3, which was derived from the EST. This approach, which has previously never been used, resulted in an increase to 84% correct classifications in the WEC test.

L2 ANSWER 3 OF 11 MEDLINE DUPLICATE 1
AN 2000260706 MEDLINE
DN 20260706 PubMed ID: 10803561
TI ECVAM's in-house prevalidation/validation studies in the areas of haematotoxicity, reproductive toxicity, metabolism-mediated toxicity and epithelial barrier function.
AU Prieto P
CS European Commission, Institute for Health and Consumer Protection, Joint Research Centre, ECVAM, Varese, Italy.. maria.prieto-pilar@jrc.it
SO SCIENCE OF THE TOTAL ENVIRONMENT, (2000 Mar 20) 247 (2-3) 349-54. Journal code: UJ0; 0330500. ISSN: 0048-9697.
CY Netherlands
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200006
ED Entered STN: 20000629
Last Updated on STN: 20000629
Entered Medline: 20000619
AB The European Centre for the Validation of Alternative Methods (ECVAM) facilitates, co-ordinates and participates in validation activities at the
European Union level. Various experimental studies, e.g. in the areas of haematotoxicity, reproductive toxicity, nephrotoxicity and epithelial barrier function, and metabolism-mediated toxicity, are underway in ECVAM's laboratories. ECVAM itself is currently involved in the prevalidation/validation of two assays, the colony-forming unit granulocyte/macrophage (CFU-GM) assays for predicting acute neutropenia and the **embryonic stem cell test**
for predicting embryotoxicity. In the areas of metabolism-mediated toxicity and nephrotoxicity and epithelial barrier function, several assays are in the course of development. In many cases, the recommendations of various ECVAM workshops are being followed.

L2 ANSWER 4 OF 11 BIOSIS COPYRIGHT 2001 BIOSIS
AN 2001:184088 BIOSIS
DN PREV200100184088
TI The use of transgenic embryonic stem (ES) cells and molecular markers of differentiation for improving the **embryonic stem cell test** (EST.
AU Spielmann, H. (1); Scholz, G. (1); Klemm, Z. M. (1)
CS (1) National Center for the Documentation and Evaluation of Alternatives to Animal Experiments at the BgVV (Federal Inst. for Health Protection of Consumers and Veterinary Medicine), Berlin Germany
SO Congenital Anomalies, (September, 2000) Vol. 40, No. 3, pp. 185-186. print.
Meeting Info.: 6th Scientific Meeting of the International Federation of Teratology Societies and the 40th Annual Meeting of the Japanese Teratology Society Matsue, Japan July 12-14, 2000
ISSN: 0914-3505.
DT Conference
LA English
S

L4 ANSWER 2 OF 25 MEDLINE
AN 2001252946 MEDLINE
DN 21146215 PubMed ID: 11248842
TI [Innovative cell culture methods in drug development]. Moglichkeit der Nutzung von Zellkulturmethoden in der Arzneimittelentwicklung.
AU Schleger C; Krebsfaenger N; Kalkuhl A; Bader R; Singer T
CS Boehringer Ingelheim Pharma KG, D-Biberach.
SO ALTEX, (2001) 18 (1) 5-8.
Journal code: DXM; 100953980. ISSN: 0946-7785.
CY Germany: Germany, Federal Republic of
DT Journal; Article; (JOURNAL ARTICLE)
LA German
FS Priority Journals
EM 200106
ED Entered STN: 20010625
Last Updated on STN: 20010625
Entered PubMed: 20010315
Entered Medline: 20010621
AB The animal studies necessary for drug registration are time-consuming, costly, and often stressful for the animals. **Toxicological** screening of drug candidates early in development with in vitro cell culture systems is therefore of relevance. In contrast to animal studies, in vitro cell culture methods are characterized by a low compound requirement and a short duration. Additionally it is possible to include mechanistic studies or to test for **toxicity** specific to humans. Therefore, early **toxicological** screening can provide a useful support for selecting the most promising drug candidate. Primary hepatocytes can be used to measure the cytotoxicity of a test compound. These results can be used to estimate general **toxicity**. Measuring endpoints like apoptosis, redox status, or gene **expression** profiles can help to answer mechanistic questions. The use of primary human hepatocytes provides early predictivity for hepatotoxicity specific to humans. Since **teratogenic** findings in animal studies often lead to abandonment of development, it is reasonable to use an in vitro embryotoxicity assay for early determination of the **teratogenic** potential of a compound, e.g. the **embryonic stem** cell test (EST) which was recently developed by ZEBET. In the EST **embryonic stem** cells are investigated for their preserved capability to differentiate into cardiomyocytes following drug exposure. In comparison cytotoxicity of the test substance is analyzed in **embryonic stem** cells and in differentiated fibroblast cells. In a validation study initiated by ECVAM the EST shows a high